IN THE CLAIMS

1-11 (Cancelled)

12. (Currently amended): A method for preparing a target protein with a carboxy~~C~~-terminal thioester, comprising:

(a) expressing in a host cell, a recombinant precursor protein ~~in a host cell, the precursor protein~~ comprising the target protein fused at its carboxy terminus to an intein, the intein having an amino-terminus and a carboxy-terminus wherein the amino-terminus is fused to the target protein and the carboxy-terminus and is optionally fused to a binding protein ~~binding~~ domain, the intein being selected from a ~~naturally occurring~~ native intein, an intein derivative or an intein mutant; ~~wherein the intein is capable of being cleaved from the protein in the presence of 2-mercaptoethanesulfonic acid~~; and

(b) contacting the expressed precursor protein with 2-mercaptoethanesulfonic acid and inducing cleavage of the intein from the precursor protein so as to form the target protein having the carboxy~~C~~-terminal thioester.

13. (Currently amended): The method according to claim 12, wherein the intein is selected from *Saccharomyces cerevisiae* ~~Sce~~ Vma intein and *Mycobacterium xenopi* ~~Mxe~~ Gyr A intein .

14. (Currently amended): The method according to claim 12, wherein the ~~protein~~ binding protein domain is a chitin binding domain.

15. (Currently amended): The method according to claim 12, wherein the target protein is selected from a *Bacillus stearothermophilus* ~~Bst~~ DNA polymerase I large fragment, thioredoxin or a cytotoxic protein.

16. (Currently amended): The method according to claim 12, wherein the binding protein domain is selected from a maltose binding protein and paramyosin.

17. (Currently amended): A method for expressing a recombinant protein precursor, comprising:

(a) inserting a nucleic acid sequence encoding a target protein into a plasmid at a multiple cloning site located upstream of and in frame with a fusion gene encoding an intein and a binding protein domain, wherein

(i) the intein is selected from a naturally occurring intein, an intein derivative or an intein mutant; and

(ii) the multiple cloning site contains a linker and the linker sequence is selected from SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3 or SEQ ID NO:4; and

(b) introducing the plasmid into a host cell and providing conditions suitable for expressing the recombinant precursor protein by the host cell.

18. (Currently amended): The method of claim 17, wherein the binding protein domain encoded by the ~~nucleic acid~~ fusion gene is a chitin binding protein.

Claims 19-20 (Cancelled)

21. (Previously added): The method according to claim 17, wherein the plasmid is a pTXB plasmid.

22. (Currently amended): A method of modifying a target protein by ligating a ~~synthetic~~ chemically synthesized peptide or ~~synthetic~~ ~~second~~ protein *in vitro* to the target protein, comprising:

(a) expressing in a host cell, the target protein fused at its carboxy terminus to ~~one of~~ an intein selected from the group consisting of: an intein, an intein derivative or a mutant intein, the intein optionally fused to a binding protein domain at its carboxy terminus, wherein the intein is capable of thiol induced cleavage;

(b) inducing ~~intein mediated~~ cleavage of the intein from the target protein by adding 2-mercaptoethanesulfonic acid so as to form a carboxy~~C~~-terminal thioester on the target protein;

(c) ~~preparing~~ obtaining the chemically synthesized ~~a synthetic~~ peptide or ~~a synthetic second~~ protein having an ~~N~~amino-terminal cysteine; and

(d) ligating the target protein of step (b) to the ~~synthetic~~ chemically synthesized ~~synthetic~~ peptide or ~~a synthetic second~~ protein of step (c) to form a modified ~~modify the~~ target protein.

23. (Currently amended): The method according to claim 22, wherein the protein after ~~prior to~~ modification is a cytotoxic protein.

24. (Cancelled)

25. (Currently amended) A method of labeling a target protein, comprising:

(a) expressing a recombinant precursor protein in a host cell, the precursor protein comprising the target protein fused at its carboxy terminus to an intein, the intein having an amino and a carboxy terminus such that the intein is fused at the amino terminus to the target protein and optionally fused to a binding protein domain at the carboxy terminus, the intein being selected from a naturally occurring intein, an intein derivative or an intein mutant, wherein the intein is capable of thiol induced cleavage;

(b) cleaving the precursor protein in the presence of 2-mercaptoethanesulfonic acid so as to form the target protein having a ~~C~~carboxy-terminal thioester;

(c) ~~preparing~~ obtaining a chemically synthesized ~~synthetic~~ peptide or protein having a marker and an ~~N~~amino-terminal cysteine; and

(d) ligating the target protein of step (b) with the chemically synthesized ~~synthetic~~ peptide or protein of step (c) for labeling the target protein.

26. (Previously amended): The method according to claim 25, wherein the marker is selected from the group consisting of a fluorescent marker, a spin label, an affinity tag, and a radiolabel.

27. (Currently amended): The method according to claim 25, wherein the chemically synthesized peptide or protein ~~fragment~~ is an antigenic determinant.

28. (Currently amended): A method for ligating a ~~synthetic~~ chemically synthesized protein or peptide to an inactive form of a protein so as to restore protein activity, comprising:

(a) expressing in a host cell, a fusion protein comprising the ~~first target~~ inactive form of the protein fused at ~~the~~ its ~~C~~carboxy-terminus to one of an intein, an intein derivative or an intein mutant wherein the fusion protein is expressed from a plasmid;

(b) inducing intein mediated cleavage of the protein of step (a) by adding 2-mercaptoethanesulfonic acid so as to form a ~~C~~carboxy-terminal thioester on the inactive protein;

(c) ~~preparing~~ obtaining a chemically synthesized ~~synthetic~~ protein or peptide having an ~~N~~amino-terminal cysteine; and

(d) ligating the inactive ~~form of the~~ protein of step (b) to the chemically synthesized ~~synthetic~~ peptide or protein of step (c) to restore protein activity.

29. (Previously amended): The method according to claim 28, wherein the protein is a cytotoxic protein.

30. (Previously amended): The method of claim 29, wherein the cytotoxic protein is a restriction endonuclease.

Claims 31-33 (Cancelled)